Mean Kinship and genetic history of the Leonberger

Dr. ir. Pieter Oliehoek - www.dogsglobal.com

There is one main reason why this report came into existence. Dog breeds are losing genetic diversity. With this loss, breeds also lose their capacity for improvement and probably more important; breeds are more prone to breed specific diseases and inbreeding depression. The bad news is: at the moment almost no breed is managed to avoid this problem. This there is only one measure taken which in itself seldom harm a breed, do also not help to save diversity:

Avoidance of inbreeding: though intuitively one might think that avoiding inbreeding will save diversity, this is not the case. Rules like forbidding brother-sister, parent-offspring mating have absolutely no influence on saving diversity. Any scientist that argues that avoidance of inbreeding *does* save diversity, does not understand conservation genetics yet. Please send them to me (dr. ir. Pieter Oliehoek).

While these measures hardly contribute in saving diversity, there is a tool that has much more potential: Mean Kinship. Mean Kinship is a value of an animal towards the current living population; it is their average (mean) relatedness (kinship) towards their population. Mean Kinship is the most important conservation tool used by zoos, to save genetic diversity within endangered species.

The breeding history of Leonberger is very similar to many other dog breeds. Therefore it was expected to have lost genetic diversity as well. To help saving diversity within the Leonberger, Mean Kinship has been calculated for the current Leonberger population.

Current Population

Besides a mean kinship of every living animal at the moment, also current and historical diversity is calculated for the Leonberger. The most important numbers, however is the diversity, kinship and inbreeding of the current population:

Founder Genome Survival   5.83
Founder Genome Equivalents 1.59
Average Mean Kinship   31.4%
Average Inbreeding   32.3%

This numbers above mean that from the original founders only 5.83 founders are left in the current population. However, because some founders and many dominant ancestors contribute much more than others, the actual diversity is about three times lower: 1.59 in 'founder genome equivalents' (or in other words: 1.59 unrelated animals. This last number can also be expressed in percentage: 31.4. This means that on average, each animal is 31.4 % related to each other. The average inbreeding is just a little higher, which is more or less what is expected.

FGE (Founder genome equivalents) is the number of founders (unrelated animals) that if bred at random would cause the same diversity (and thus average mean kinship) as the current population. In the Leonberger this is 1.59. The overall diversity is thus lower than if you would start the same population with 2 animals. The average mean kinship is therefore higher than a brother-sister-mating (which would be 25%).

Genetic History of the breed

Founders

Most dog breed populations have a history not more than 100 years old. For the Leonberger this is slightly older. Because of the extensive dataset available it is possible to identify the founders: unrelated animals that started the population. Every gene that is present in the current population must descent from one of these founders. The algorithm found 14 founder-animals contributed to the current population.

Name Founders	Sex	Born	Pups	Rep	%
Lord v. Langen	M	1911	2	2	6.6
Senta v. Langen	F	1911	2	2	6.6
Leo E4	M	1912	16	3	4.1
Bella v.d. Kochlinsmuhle	F	1914	5	4	14.7
Marco v.d. Kochlinsmuhle	M	1914	5	4	14.7
Minka v. Karlsruhe	F	1914	1	1	3.5
Minka v. Reihen	F	1915	1	1	7.1
Frika v.d. Mussigmuhle	F	1918	19	3	4.1
Marko v. Schwaigern	M	1920	16	5	19.2
Flora II v. Kochertal	F	1921	3	2	14.7
Leonora v.d. Schwarzach	F	1921	10	1	2.5
Treu v. Kufstein	M	1922	17	1	0.8
Grisette v. Bruckberg (Newfoundlander)	F	1947	1	1	1.2
Telu (a.k.a. Tetu) Non Leonberger	F	1972	2	1	0.1
				Total:	100.0

Name Foundlings	Sex	Born	Pups	Rep	%
Asta v. Tachenhausen DLZB 1450	F	1944	15	4	5.1
Tasso OHZB L 42	M	1955	6	1	4.0
				Total:	9.0

"**Pups**", means: the number of progeny (puppies) a founder had. "**Rep**" is the number of puppies that actually reproduced themselves. Four founders contribute more than half to the entire current population.

Besides founders, there were also two animals found from which no parents were known, however, after historical research we believe they were not founders. Their parents were most likely related, but we simply don't know who they were. These animals are called 'foundlings'. Two foundlings were identified with high impact to the current population. Their contribution to the current population is 9%. With the calculation of kinship, founder genome surviving and inbreeding levels, an algorithm make sure these animals were not seen as unrelated, which would have influenced the results dramatically. The algorithm used was the C3-algorithm of chapter 4 from the thesis: "Genetic Conservation of Small Animal Populations" (Pieter Oliehoek 2009). This thesis can be found here: http://www.breedingfordiversity.com/thesis/ This thesis describes much more about foundlings and genetic diversity as well.

Recently, the Average inbreeding was calculated also by the University from Bern, which resulted in an average of 29%. This number is about 3% lower than the inbreeding found here: 32%. This lower number in the Bern-research is most likely due to the foundlings being regarded as founders.

Dominant Ancestors

Some animals contributed much more than others. This is not only because they had a lot of progeny, but can also be because their progeny had a lot of progeny, and so on. Through the generations, some animals can have a very dramatic impact on an entire populations. The following table shows the ancestors that contributed most to the current population.

Name	Sex	Born	Father	Mother	Pups	Rep	%
Asta v. Romerturm	F	1922	Marko v. Schwaigern	Flora II v. Kochertal	31	6	22.8
Cora v. Wintersheim	F	1940	Wotan v. Wintersheim 773	Adda v.d. Harmonie	9	3	22.4
Marko v. Neukirch	M	1920	Marco v.d. Kochlinsmuhle	Bella v.d. Kochlinsmuhle	81	9	20.5
Leo v. Stern	M	1938	Marko v. Leonberg	Leda v. Jugoslavien	9	5	20.0
Ali v. Martinsfeld	F	1944	Leo v. Stern	Toni v.d. Dune	15	5	18.4
Casar v. Wintersheim 1209	M	1944	Leo v.d. Dune	Cora v. Wintersheim	39	7	18.3
Arko v. Leonberg	M	1949	Casar v. Wintersheim 1209	Birka v. Carolshofen 1461	103	8	17.9
Carlo v. Glemstal	M	1955	Arko v. Leonberg	Ali v. Hohenreutin	110	13	17.8
Ajax v. Nussloch	M	1933	Astor v. Uhingen	Alma v. Nussloch	87	9	17.2
Baldur v. Wintersheim 1147	M	1942	Jokel v. Bendstich	Cora v. Wintersheim	61	5	16.3
Birka v. Carolshofen 1461	F	1947	Baldur v. Wintersheim 1147	Ali v. Martinsfeld	15	3	15.3
Leo v.d. Sagmuhle	M	1922	Nero v.d. Elsenz	Senta v. Wintersheim DLZB 69	44	7	14.6
Marko v. Leonberg	M	1933	Eberhard v. Schwarzwaldrand	Addy v. Rottweil	23	2	14.4
Nero v.d. Elsenz	M	1919	Leo v. Ziegelhof	Minka v. Reihen	27	6	14.1
Eberhard v. Schwarzwaldrand	M	1927	Arno v. Schwarzwaldrand 153	Asta v. Romerturm	21	4	13.7
Jokel v. Bendstich	M	1939	Cero v. Stern	Circe v. Bendstich	4	2	13.2
Adda v.d. Harmonie	F	1933	Bub v. Schwarzwaldrand	Bella v. Oftersheim	13	3	12.8
Wotan v. Wintersheim 773	M	1934	Alf v. Aldingen	Senta v.d. Elsenz	6	2	12.2
Barko v. Hohen Karpfen	M	1954	Arko v. Pappelhof	Betty v. Carolshofen	79	11	11.7
Baldur v. Leonberg	M	1950	Ortwin v. Bendstich	Birka v. Carolshofen 1461	77	10	11.6
Ortwin v. Bendstich	M	1947	Abel v. Heidekaten	Kuni v. Bendstich	78	7	10.8
Addy v. Rottweil	F	1930	Leo v. Hailtingen	Cora v. Schwarzwaldrand 247	14	3	10.7
Bella v. Oftersheim	F	1931	Astor v.d. Dune	Alma II v.d. Krone	8	3	10.6
Leo v.d. Dune	M	1939	Ajax v. Nussloch	Alli v.d. Drei Eichen	13	2	10.5
Arko v. Pappelhof	M	1948	Barras v. Wintersheim	Addy v. Carolshofen	34	4	10.4
Alli v.d. Drei Eichen	F	1933	Benno v.d. Elsenz	Flora v. Schwarzwaldrand	15	2	10.0
Leda v. Jugoslavien	F	1937	Bar v. Schwarzwaldrand	Addy v. Aldingen	3	1	10.0
Senta v. Wintersheim DLZB 69	F	1921	Lord v. Langen	Senta v. Langen	5	3	9.7
Barras v. Wintersheim	M	1942	Jokel v. Bendstich	Cora v. Wintersheim	15	4	9.4
Addy v. Carolshofen	F	1946	Leo v. Stern	Ali v. Martinsfeld	12	4	9.4
Afra v. Schwarzwaldrand	F	1924	Marko v. Neukirch	Asta v. Romerturm	19	5	9.4
Toni v.d. Dune	F	1938	Ajax v. Nussloch	Alli v.d. Drei Eichen	10	1	9.2
Ali v. Hohenreutin	F	1950	Argus v. Pappelhof	Asta v. Tachenhausen DLZB 1480	18	1	8.9
Cero v. Stern	M	1937	Marko v. Leonberg	Betty v.d. Harmonie	26	3	8.9
Astor v.d. Dune	M	1929	Lord v. Wimpfen	Bella v.d. Krone	10	4	8.8
Astor v. Uhingen	M	1928	Castor v.d. Karlsaue	Asta v. Hohenberg	12	1	8.6
Alma v. Nussloch	F	1931	Achill v.d. Dune	Bella v. Wildberg	6	1	8.6
Bella v.d. Krone	F	1926	Leo v.d. Sagmuhle	Afra v. Schwarzwaldrand	10	3	8.5
Bub v. Schwarzwaldrand	M	1931	Arno v. Schwarzwaldrand 448	Alma v. Schwarzwaldrand	5	2	8.4
Arko v. Gaisberg	M	1966	Boris v. Staufenzwinger	Diana v. Glemstal	55	12	8.0
Alma v.d. Niederhaid	F	1957	Tasso OHZB L 42	Dorle v. Nussbaum	19	8	7.9
Minka v.d. Elsenz 385	F	1925	Leo v. Hillisheim	Anita v. Glemstal	12	3	7.8
Arras v. Krametsbuhl	M	1969	Gyurcsitarjan Azor	Freya v. Murrtal	195	31	7.7
Gyurcsitarjan Iwanowitsch	M	1971	Gyurcsitarjan Zegty Zorvath Zwo	Alkeste v.d. Danubia	182	19	7.6
Gyurcsitarjan Azor	M	1963	Alf v.d. Achalm	Elka v. Rossbach	77	11	7.4
Argus v. Pappelhof	M	1948	Barras v. Wintersheim	Addy v. Carolshofen	26	5	7.3
Benno v.d. Elsenz	M	1927	Marko v. Schwaigern	Minka v.d. Elsenz 385	30	2	7.2
Bella v. Tivoli	F	1952	Alex v. Carolshofen	Astra v. Saul	27	6	7.2
Alma v. Schwarzwaldrand	F	1924	Marko v. Neukirch	Asta v. Romerturm	2	2	7.1
Arno v. Schwarzwaldrand 448	M	1928	Leo v.d. Sagmuhle	Diana v.d. Schwarzach	14	2	7.1
Lord v.d. Solitude	M	1962	Carlo v. Glemstal	Cora v.d. Solitude	57	12	7.1
Leo v. Ziegelhof	M	1916	Leo v. Karlsruhe	Minka v. Karlsruhe	1	1	7.1

Two ancestors almost contributed one fourth to the entire current population. Their impact is very high on the relatedness within the population (making all animals family), the inbreeding due to this relatedness and most likely on inbreeding depression that is very likely to follow. Furthermore, since every animal (and human) will carry at least one recessive genetic disease with them, the specific recessive diseases that these animals were carrying are now spread throughout the entire population. To lesser extend this is also true for every other animal on the dominant ancestor list.

Average Mean Kinship and Inbreeding over time

The following graph shows the average mean *kinship* and the *inbreeding* in percentages over the known pedigreed history of the Leonberger. Though most focus often is inbreeding, average mean kinship is actually more important, since inbreeding is caused by kinship and not opposite.
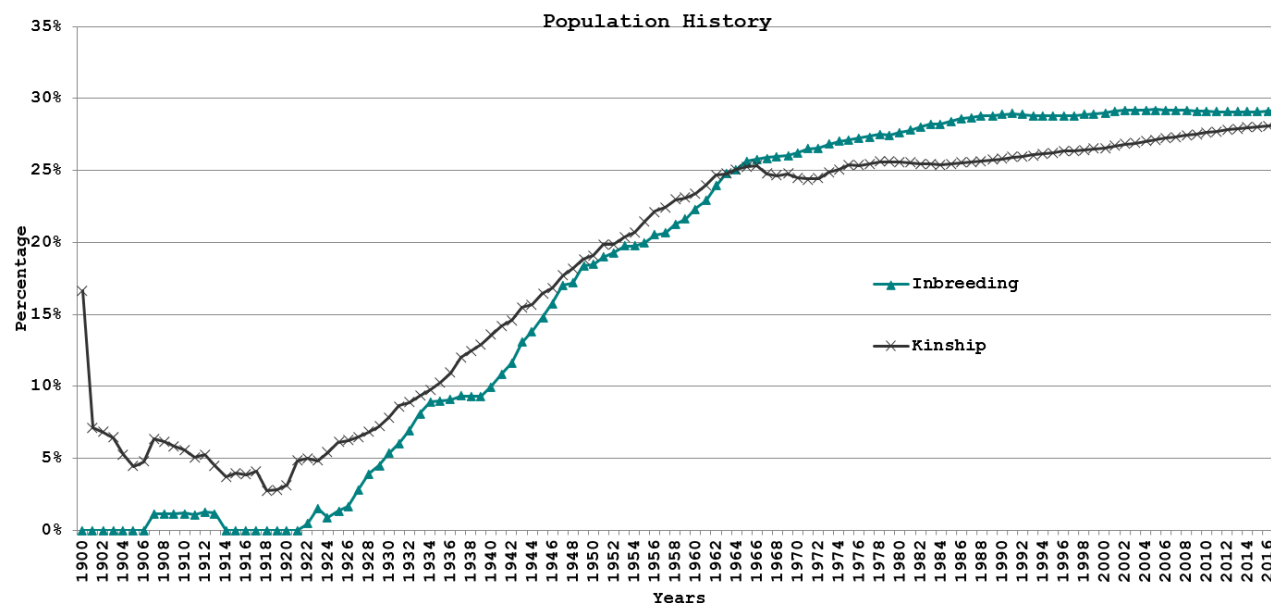
Before the 70s, the population size in the Leonberger was much lower. From the 70s on the number of litters increased largely. There are now over 4000 animals born per year. This does not necessarily mean that a populations keeps its diversity, or in other words, does not become more and more related and therefore inbred. In fact, during the last decades it is obvious that the kinship (relatedness) of the populations gradually keeps on climbing. If nothing changes in breeding strategies, it is expected that this increase of kinship (and thus increase of inbreeding) will continue.

Before the 60s, inbreeding behaved as expected: following the kinship. After however, it started to be higher than kinship. Nowadays however they are almost at the same level again. Since inbreeding follows kinship, it is expected that inbreeding will go up together with the kinship. If you would only look at the inbreeding level, one might wrongly assume that the problem is not that high in recent years. Note that avoidance of inbreeding does not save diversity, whatsoever. Inbreeding is a result. This cannot be pointed out often enough. The main goal for a population is to keep diversity high and thus kinship low. And the graph shows a steady increase. Because inbreeding is now close to the level of kinship, the relative low increase of inbreeding since the nineties will now cease and soon the inbreeding will go up, following the increase of (average mean) kinship.

For the biologists among you: inbreeding could also be interpreted as "observed homozygosity" while average mean kinship is the "expected homozygosity". Genetic diversity is then the "expected heterozygosity" and the Founder Genome Equivalents is literally the equivalents in the number of founders having such an expected heterozygosity.

Mean Kinship

Mean kinship was calculated for every animal of the current population. The current population was estimated to be about 33.000 animals. The current population is determined from the database and includes all individuals that are at most 9 years old and not known to have died. The actual population size might therefore be smaller, since for many animals we do not know their current status or will never breed. Animals older than 9 years were presumed not to participate in breeding anymore.

From the pedigree data kinships were calculated up to the founders among all living individuals. After calculating mean kinship (MK) for each animal, the population was separated into three groups: animals having low mean kinship (the green list), animals having high mean kinship (the red list) and the animals in between: (the yellow list).

Green: MK values till 0.32
Yellow: MK values between 0.32 and 0.3285
Red: MK values from 0.3285 onwards
Note that these values are arbitrary and solely chosen to split the population into three equal groups.

The following table shows animals by list and by age group:

Age	Green	Yellow	Red
0		21	13
1	732	1059	967
2	974	1366	1430
3	1034	1507	1373
4	1267	1538	1423
5	1203	1618	1456
6	1449	1479	1343
7	1475	1505	1272
8	898	756	575
9	835	726	538
Total:	9867	11554	10377

This table is interesting, because it can slightly predict what is going to happen in the future. The younger animals will take over the breeding of the older animals. The red list contain relatively more young animals while the green list contain relatively older animals. If breeding would continue without looking at mean kinship, and the young animals are taking over, most breeding will be done with animals from the red list. This will for sure lead to an increase of the average mean kinship as well. Within the Icelandic Sheepdog we found families that were highly unrelated to the overall populations. We hoped to find the same within the Leonberger population, alas this was not the case.

Sometimes people assume that each country has its own population and individuals within the country are more related to each other than between countries. In practice this might not be the case at all. There is no saying how diversity is spread over countries, unless you calculate this. In the end there is only ONE Leonberger population. All countries share the responsibility to maintain the diversity of the Leonberger as a whole. The following table shows the spread of animals from each list for each country.

Country	Green	Yellow	Red
Australia	23	63	54
Austria	95	74	
Belgium	365	639	294
Canada	90	554	305
Czech Republic	492	538	218
Denmark	114	140	96
Estonia		21	79
Finland	37	474	1277
France	6096	1378	227
Germany	677	1951	1681
Great Britain	371	1116	745
Hungary	35	274	203
Ireland	14	18	60
Italy	127	589	419
Japan			47
Latvia	18	43	26
Netherlands	514	956	511
New Zealand	90	46	204
Norway		76	656
Other	2	41	50
Poland	316	426	135
Russia	4	188	230
Slovak Rep.	36	126	38
Spain	91	46	12
Sweden	20	213	2022
Switzerland	89	168	56
USA	151	1417	745
Total:	9867	11575	10390

Breeding is done preferably with animals from the green list. This will increase the diversity within the population. Ignoring these animals would result in the opposite: the genetic diversity will continue to go down. The table clearly shows that some countries have much more red-list animals while others have more green-list animals. Though the reason behind this might be due to differences in breeding policies, this is extremely difficult to be proven. It is much more important to understand that without these MK-calculations it is impossible to know if an animal is unrelated to a population or not. This is at the same time the reason Mean Kinship was developed in the first place. Zoos having dramatically small populations, could not distinguish the related from the unrelated from pedigree, though the studbook-keepers are working with the populations on daily basis. In 2003 the Icelandic Sheepdog population was also analysed and could make use of Mean Kinship. The Leonberger is the second dog population in the world that is able to use Mean Kinship to increase the genetic diversity and ultimately decrease inbreeding levels. Till now no one could have known which countries would have most unrelated animals.

Recommendations

It is clear that the diversity in the Leonberger is very low. The numbers show high levels of (average mean) kinship and inbreeding. If current trends continue, which will be the case if same breeding policies are applied, the kinship will also continue to rise. Inbreeding is expected to increase even a little faster.

Furthermore, the population does not contain 'hidden families' that are unrelated to the majority of the population. Instead the Leonberger is one large family with little differences in kinship. There is no two animals in this population with a lower relation than at the level of full-siblings (brother and sister mating).

However, there IS still diversity present within the Leonberger that could be used to increase the diversity of the population a little. As previously described, these animals need to be used and preferably quickly, because most of the green-list animals are older in age. This would be the smart way forward: start focussing on animals that can contribute to this diversity, mainly the green-list animals. These animals are not equally spread over each country. Per country, therefore another strategy would be recommended. To breed for diversity it is not necessarily best to import from other countries. In fact, in some cases it could even diminish potential genetic diversity, which has been the case within the Icelandic Sheepdog. The most important thing is not to focus on countries, but on green-list-animals, where-ever they are.

The majority of diversity is present in France. For France it is therefore not recommended to import animals from other countries. This does not mean that France can 'relax'. Also within France a focus on the green-list-animals would help the diversity of the breed. Further recommendations are simply following common sense. For none of the countries is it advisable to import animals from Norway, Sweden, Finland, Russia, Japan, USA or New Zealand and other countries that have hardly any green-list animals present. Especially for those countries, it would be interesting to import green-list animals from France, since the majority of the green-list animals can be found there.

Even for countries that mainly have green-list animals, like Austria, Spain or Poland, it would still help to import from France if these animals come from the red-list. For them, the necessity is of course less high.

Mean Kinship is a tool that provides us insight and opportunity to maintain the genetic diversity of our breed and even (although slightly) improve the overall genetic diversity of the population, which is the best way to avoid future health problems of a breed. Though Mean Kinship is a tool to help the diversity of the breed, it does not help the current health of the breed, nor does it help the 'diversity of a litter'. Hence, Mean Kinship is not a replacement for other tools. It is still important to avoid inbreeding, especially between close family members. For example even though a dam and sire are both in the green zone, does not necessarily mean they are a good match. There is a small chance that these two dogs themselves could be closely related, for example a full brother and sister will both be in the same zone! The same would be true for the red zone or the yellow zone of course. But in most cases even green bitches are preferred to be bred with dogs from the green zone as well.

Besides focussing on the green zone, it is also important to limit the number of litters per dog and check for genetic diseases that have strong implications for suffering of the puppies and are already highly spread throughout the population. Since there are no separate unrelated families in the Leonberger, it is highly likely that breed specific diseases are not bound to specific families or lines, but probably spread throughout the population. Mean Kinship can help to avoid that new disease become prevalent as well, but in the case of the Leonberger cannot aid in getting rid of genetic diseases that are now already there.

To transpose the research results into breeding policies is a delicate and complex matter. In my experience, scientific findings are too often abused for people's own agenda. For this reason I conclude this report with a warning. Conclusions based on these results should not be taken lightly.

There might be an inclination to use this report to claim that the Leonberger has a problem, or that the Leonberger does not have a problem. Both conclusions cannot be confirmed by this report. The only conclusions that are valid in the author's opinion are (in 2018):

1) the Leonberger did lose a dramatic proportion of its original genetic diversity
2) it is likely that this loss will continue if breeding continues in the same way
3) there is space to increase / restore some of the genetic diversity that was lost without having to use other breeds, by means of using the green list animals
4) when importing from other countries, the best source for green-list animals of France

Data in this report should not be published elsewhere without permission of P.A Oliehoek, with the exception for direct communication towards breeders of the Leonberger.

About the author

Dr. ir. Pieter (P.A.) Oliehoek has been in involved in dog breeding since he was eleven years old. Because of his love for carnivores he studied biology at the International University of Wageningen (WUR). During his Msc he did an internship on separating dog breeds with the use of DNA and did his thesis on conservation of the Icelandic Sheepdog with the aid of pedigree-data and cluster analysis. Pieter was involved in the early years of the foundation of the Icelandic Sheepdog International Committee (ISIC - www.icelanddog.org). After three years of working as an ICT professional, he went back to the university to investigate how to genetically preserve rare livestock breeds and endangered species in captivity. He received his PhD on this topic in 2009 at the WUR. Since then Pieter is involved on projects within zoo populations on how to apply complicated theory in day to day practice. His scientific work can also be found on: www.breedingfordiversity.com. Since 2018, he started to concentrate his full effort again on dog breeding. Pieter is the founder of **www.dogsglobal.com** an informative website with the goal to increase health of all dogs in general and dog breeds in particular by sharing information.

DOGS GLOBAL